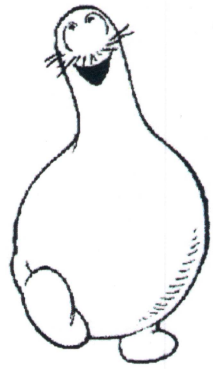

Colony PCR for Screening Yeast

1. Pick a single colony using a wooden stick or inoculating loop and patch on a dropout plate. Take stick and rub into a PCR tube with 50 µl of Zymolyase working solution (60U/mL. Stock solution is 5U/µl, to make working solution take 12 µl stock and add to 988µl of water).

2. Incubate at 37 degrees for 30 minutes, then 95 degrees for 10 minutes.

2. Set up PCR reaction as below:

	1X reaction
2X GoTaq Green PCR Master Mix	10 µl
10 µM FW primer	1 µl
10 µM REV primer	1 µl
water	3 µl
boiled yeast cells (template)	5 µl

Cycles:

95		5 min
30x:	95	45 s
	50	30 s
	72	1 min per kb
72		10 min

3. Analyze products on a 1% agarose gel. The GoTaq mix already has gel loading dye in it, so you can just directly load 5 µl of your PCR reaction into a gel.